

veronal caused the cytoplasm to appear more homogeneous. The resulting micrographs most closely resembled those Porter had produced with tissue-cultured cells. Palade's buffered solution of 1–2%  $\text{OsO}_4$  in a neutral Michaelis buffer became known as “Palade's Pickle” and was widely adopted. Dalton (1955) added chromium salts to cross-link proteins and stabilize them.

Although Palade's technique for staining cells with  $\text{OsO}_4$  was widely adopted, that did not mean that there were no controversies over its application. Sjöstrand took issue with Palade on several aspects of the technique. For example, while faulting the time lag in Palade's application of the fixative (which, he claimed, caused tissues to deteriorate), Sjöstrand also dismissed the importance Palade had placed on maintaining the pH of the specimen:

We owe a good deal to Palade (1952) who modified the osmium fixation to give a decent preservation of the fine structure of cells. His introduction of the veronalacetate buffer as a medium for the osmium tetroxide has improved the quality of fixation for several tissues. However, the pH of the osmium tetroxide seems not to be so important as he originally claimed. Variation of pH within a wide range from pH 4, and in some cases even pH 2, to pH 8 does not substantially affect the result in many tissues, e.g., the kidney (Rhodin, 1954), the intestinal epithelium (Zetterqvist, 1956) and the sensory epithelium of the inner ear (Wersäll, 1956). Most of the great differences of the quality of fixation described by Palade (1952) in his study at a low resolution seem to represent various degrees of post-mortem changes. (1956a, p. 459)

To speed fixation, Sjöstrand injected fixative into live animals and immersed cut tissue in fixative as quickly as possible. He also claimed that the tonicity of the fixative, which Palade argued was not critical, was very important in preventing swelling:

The osmium tetroxide solution of Palade is strongly hypotonic and in several cases . . . hypotonic solutions of osmium tetroxide produce a more or less marked swelling of the cells. The importance of using isotonic solutions has therefore been stressed (Sjöstrand, 1953a). It has even been possible to obtain as good a preservation of the cell structure by using isotonic non-buffered solutions of osmium tetroxide as for instance a solution of isotonic sodium chloride. (Sjöstrand, 1956a, p. 459)

Rasmussen traced the methodological dispute between Palade and Sjöstrand in part to a difference in the use to which they put electron micrographs. For Palade, the use was primarily qualitative – to provide morphological perspective on the biochemical information generated, for example, from fractionation and functional analysis of mitochondria and mitochondrial fragments.